ABSTRACT

Microbes isolated from diverse environmental sources such as contaminated food, nitrogen rich soil, tannery wastewaters, soils from high altitude, activated sludges from effluent treatment plants dealing with pesticide and oil refineries were found to belong to Bacillus, Bordetella, Anoxybacillus, Myroides, Alcaligenes, Marinobacter, Halomonas, Proteus and Pseudomonas sp. on the basis of 16S rRNA gene sequence analysis. More than hundred microbial isolates were screened out of which 60 isolates had the ability to produce polyhydroxybutyrate (PHB) from 15 to 565 mg/liter of medium, equivalent to a yield of 2 to 67%. The highest PHB yield of 67% was recorded with Bacillus cereus strain EGU3. Six selected strains were tested for their abilities to produce polyhydroxybutyrate (PHB) from different sugars and biowaste (Pea-shells). These Bacillus strains with the ability to grow on different sugars (1% w/v) were shown to produce PHB from GM2 medium supplemented at the rate of 1% (w/v) with glucose (up to 435 mg PHB/L; 31 to 62% w/w), fructose (up to 385 mg PHB/L; 2 to 75% w/w), maltose (up to 220 mg PHB/L; 12 to 48% w/w), sucrose (up to 170 mg PHB/L; 12 to 35% w/w), however, no PHB production could be detected on lactose. Out of these 6 Bacillus strains, Bacillus cereus EGU44 and B. thuringiensis EGU45 could convert a combination of biowaste (pea-shell slurry, 2% Total Solids) and GM2 medium (in 1:1 ratio, v/v) supplemented with casein enzyme hydrolysate (as additional nitrogen source) in to 945 to 1205 mg PHB/L (53 to 65% w/w). Optimization for different parameters revealed that an incubation period of 72 h, nitrogen source supplement at the rate of 0.04% and inoculum size of 100 μg cell protein/ml of biowaste : medium (BW:M :: 1:1) resulted in further improvement with a final production of 3010 to 3370 mg PHB/L, equivalent to 300 g/Kg biowaste (dry weight). This is the first report on usage of pea-shells as feed for PHB production. Six distinct strains produced polyhydroxybutyrate (PHB) on GM2 medium supplemented with 5 different sugars at the rate of 1% (w/v): up to 87% w/w on glucose, up to 50% w/w on fructose, up to 34% w/w on maltose, up to 36% w/w on sucrose and up to 54 % w/w on lactose. Biowaste, BW:medium, M :: 1:1 with 0.4 g/L casein enzyme hydrolysate supplementation was optimized previously for PHB production with pea shells as feed material.
The uniqueness of the strains tested in the last phase of experimentation lies in the fact that these strains have not been reported for PHB production so far. Six distinct strains produced polyhydroxybutyrate (PHB) on GM2 medium supplemented with 5 different sugars at the rate of 1% (w/v): up to 87% w/w on glucose, up to 50% w/w on fructose, up to 34% w/w on maltose, up to 36% w/w on sucrose and up to 54% w/w on lactose. Biowaste, BW:medium, M :: 1:1 with 0.4 g/L casein enzyme hydrolysate supplementation was optimized previously for PHB production with pea shells as feed material. Out of these 6 strains tested again on biowaste for PHB production, the highest producers were *Alcanivorax* sp. EGU619 and *Myroides odoratus* EGU882 that could produce PHB to the level of 1015 and 1145 mg PHB/L respectively at 96h.